

## **Standard Operating Procedure for the Determination of Dissolved Oxygen by the Membrane Electrode Method and Modified Winkler Method**

### **1.0 Location**

These methods are performed in the BOD area of room 305.

### **2.0 Purpose**

These methods are used to determine the amount of dissolved oxygen in a liquid sample.

### **3.0 Scope**

The modified Winkler method is used to determine the amount of dissolved oxygen in dilution water. The value obtained is used to calibrate the DO probe for BOD determinations.

### **4.0 References**

4.1 Standard Methods for the Examination of Water and Wastewater, 17th Edition, p.4-149 to 4-161, Method 4500-0, p 4-48 to 4-50, Method 4500-C1B.

4.2 Instruction Manual for Orion Oxygen Electrode, Model 97-08, 1989.

### **5.0 Sample Preservation and Handling**

This procedure is used in the laboratory to measure dissolved oxygen in diluted samples before and after incubation in the Biochemical Oxygen Demand procedure. See section 5.0 of method 9.3.

### **6.0 Apparatus and Materials**

6.1 Orion Oxygen Electrode, Model 97-08

6.2 BOD bottles, 300 ml  $\pm$  3 ml capacity.

6.3 Dilution H<sub>2</sub>O: Aerate reagent grade water with a resistivity of >10 megaohm-cm for approximately 4 hours.

- 6.4 Manganous sulfate solution: 364 g Manganous Sulfate ( $\text{MnSO}_4 \cdot \text{H}_2\text{O}$ ) in deionized water and dilute to 1 L.
- 6.5 Alkaline iodide-azide solution: Dissolve 350 g of potassium hydroxide (KOH) and 75 g of potassium iodide (KI) in chilled deionized water and dilute to 500 ml. To this solution add 5 g sodium azide dissolved in 20 ml of deionized water. It is in the little refrigerator)
- 6.6 Sodium thiosulfate, stock solution, 0.75N: Dissolve 186.15 g sodium thiosulfate 5-hydrate in deionized water and dilute to 1 liter. Refrigerate. ( At this time, we order it already made. Can be kept in cabinet until opened, then refrigerate)
- 6.7 Sodium thiosulfate standard titrant, 0.0375N: Prepare by diluting 50 ml of stock solution to 1 liter. Standardize and refrigerate. See section 7.1.
- 6.8 Starch: Prepare an emulsion of 10 g of soluble starch in a beaker or weigh boat with a small quantity of cold deionized water. Pour this emulsion into 1 liter of boiling water in a beaker and allow to boil for a few minutes. Let settle overnight. Remove scum from top and use the clear solution. Refrigerate when not in use.
- 6.9 Potassium dichromate,  $\text{K}_2\text{Cr}_2\text{O}_7$ , 0.025 N: For standardization of 0.0375N sodium thiosulfate. Dissolve 1.226 grams of dried  $\text{K}_2\text{Cr}_2\text{O}_7$  (1 hr at 105° C) in deionized water and dilute to 1L. Store in brown bottle and refrigerate. Make every 3-6 months.
- 7.0 Procedure
- 7.1 To standardize 0.0375N sodium thiosulfate.
- 7.1.1 Weigh 2.0 g KI and put in a 1L wide mouth flask. Dissolve with 150 ml of deionized water. Add 10 ml of 1:10  $\text{H}_2\text{SO}_4$ . Add 20 ml of  $\text{K}_2\text{Cr}_2\text{O}_7$  and place in the dark for 5 minutes. Bring to volume of 400 ml with deionized water. Add 1 ml of starch and titrate with 0.0375N sodium thiosulfate solution from blue to clear. Run in triplicate. Triplicates should agree within  $\pm 0.05\text{ml}$  and should be within 0.0002 of 0.0375.
- 7.1.2 N of Sodium thiosulfate =  
$$\frac{(\text{ml of } \text{K}_2\text{Cr}_2\text{O}_7)(\text{N of } \text{K}_2\text{Cr}_2\text{O}_7)}{\text{ml of titrated sodium thiosulfate}}$$
- 7.1.3 Document the standardization in a permanent record.

## 7.2 Modified Winkler Method

- 7.2.1 Prepare four BOD bottles full of dilution water. Two bottles are for calibration of the DO probes. Read the DO of each bottle and record.
- 7.2.2 To each of two bottles, add 2 ml of the manganous sulfate solution followed by 2 ml of the alkaline iodide-azide solution well below the surface of the liquid. Stopper with care to exclude air bubble and mix well by inverting the bottle several times. When the precipitate settles, leaving a clean supernatant above the manganese hydroxide flock, mix again by gentle inversion. When settling has produced at least 200 ml of clear supernatant, carefully remove the stopper and immediately add 2 ml of concentrated  $H_2SO_4$  by allowing the acid to run down the neck of the bottle, re-stopper, and mix by gentle inversion until the iodine is uniformly distributed throughout the bottle. Complete the analysis within 45 minutes.
- 7.2.3 Record beginning and ending levels in buret.
- 7.2.4 Transfer the entire bottle contents by inversion into a 1000 ml wide mouth flask and titrate with 0.0375N thiosulfate solution to pale straw color. Add 1 ml of starch solution and continue to titrate to the first disappearance of the blue color.
- 7.2.5 Each ml of the 0.0375N sodium thiosulfate titrant is equivalent to 1 mg DO.
- 7.2.6 Average the two results and adjust the DO electrode in the third bottle. See section 7.3.
- 7.2.7 Record all the values and calculations on sheet in holder near the electrode.

## 7.3 Calibrating the electrode

- 7.3.1 With the electrode mode switch in the OFF position, switch the meter to the pH mode. The reading will be 7.00.
- 7.3.2 Turn the mode switch on the electrode to BATT. Good battery operation is indicated by a reading >13.000.

- 7.3.3 Turn the mode switch on the electrode to ZERO. Use the zero calibration control to set the meter to read 0.00.
- 7.3.4 Insert the funnel into a BOD bottle full of dilution water and slowly immerse the electrode into the funnel.
- 7.3.5 Place the bottle on a magnetic stirrer and stir gently.
- 7.3.6 Turn the mode switch on the electrode to H<sub>2</sub>O.
- 7.3.7 Turn the right hand knob on the electrode to adjust the meter to read the value obtained with the Modified Winkler method.
- 7.3.8 Store the electrode in a BOD bottle containing enough water to cover the bottom but not contacting the funnel.
- 7.3.9 Record battery check, zero, bottle readings, buret readings, initials, and bottle number of H<sub>2</sub>O used on sheet in holder near electrode.
- 7.3.10 Stopper and water seal bottle used for calibration for later re-calibration as needed.
- 7.4 Analysis
  - 7.4.1 Insert the funnel into the sample bottle. Slowly immerse the electrode into the funnel.
  - 7.4.2 Place the bottle on a magnetic stirrer and stir gently.
  - 7.4.3 Turn the mode switch of the electrode to the H<sub>2</sub>O position.
  - 7.4.4 Obtain and record stable reading. The result displayed is ppm O<sub>2</sub>.
  - 7.4.5 Slowly remove the electrode from the funnel.
  - 7.4.6 Remove the funnel and insert into next sample bottle. Insert electrode. Repeat for each sample.
  - 7.4.7 Rinse electrode and funnel between samples but not between dilutions of the same sample.

7.4.8 After reading on the last sample has been taken, rinse electrode and funnel with distilled water. Gently blot membrane dry.

7.4.9 Place funnel and electrode in storage bottle.

## 7.5 Care of electrode

7.5.1 After use, rinse electrode and funnel with distilled water. Gently blot membrane dry.

7.5.2 Always store the electrode in a BOD bottle containing enough water to cover the bottom but not touching the electrode membrane.

7.5.3 In case of problems with electrode refer to trouble shooting checklist below:

## TROUBLESHOOTING CHECK LIST

<b>symptom</b>	<b>probable cause</b>	<b>solution</b>
drift or slow response	internal pressure in membrane/electrolyte module because the membrane/electrolyte module has been installed too quickly	partially unscrew the module and retighten slowly
	new module not allowed to equilibrate long enough	allow newly installed module to equilibrate for 15 to 30 minutes before calibrating
	water droplets clinging to membrane during air standardization	blot tip with tissue and recalibrate
	no stirring when electrode is in solution	make sure that magnetic stirrer is turned on and that bar is in synchronization
	batteries weak	check and replace if necessary
unable to calibrate	electrode not plugged in or meter not in pH mode	check electrode plugs and meter mode switch
	zero setting improperly adjusted	check and reset if necessary
	tip of electrode wet	blot dry
	batteries weak	check and replace if necessary

membrane/electrolyte module damaged or      replace module  
dried out

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7.5.4 If problems persist clean electrode tip and replace membrane module.

7.5.4.1 Remove membrane module from electrode.

7.5.4.2 Do not touch the anode/cathode tip with your bare fingers when membrane module is removed.

7.5.4.3 Rinse tip with distilled water.

7.5.4.4 Soak electrode tip only in 0.1M HCl (1ml HCl/120ml) for 30-60 minutes.

7.5.4.5 Rinse thoroughly with distilled water and gently blot dry. Do not wipe or rub tip.

7.5.4.6 Replace membrane module. Do not spill or remove liquid gel from inside module. Hold module upright and slowly push anode/cathode tip into it. Slowly screw module onto the electrode until it is finger tight. Place electrode in storage bottle and wait 30-60 minutes before calibrating or using.

7.5.4.7 Cleaning and new membrane module should last 3-6 months.

7.6 Checking electrode operation

7.6.1 Calibrate electrode using regular procedure.

7.6.2 Prepare a 5% sodium sulfite solution by dissolving about 12.5 g sodium sulfite in 250ml distilled water. Transfer solution to a BOD sample bottle.

7.6.3 Place the overflow funnel in the bottle, making sure that the funnel is snugly seated. Immerse the electrode in the funnel. Place the bottle on a magnetic stirrer and stir gently. Turn the mode switch to H<sub>2</sub>O.

7.6.4 After 2 1/2 minutes, the reading should be 0.1ppm oxygen or less. If not, see sections 7.5.3 and 7.5.4.

7.6.5 Slowly remove electrode and then the funnel. Thoroughly rinse each separately under cool running tap water to remove all traces of sulfite.

Rinse with distilled water.

7.6.6 Place electrode in storage bottle.

## 7.7 Measuring hints

7.7.1 To avoid spilling sample solution, always remove the electrode from the funnel before removing the funnel from the BOD bottle.

7.7.2 Slowly insert and remove the electrode from the funnel. Rapid insertion and removal may shift the electrode's standardization.

7.7.3 Keep the electrode away from any sharp objects that could puncture the membrane.

7.7.4 Thoroughly rinse the electrode and funnel separately under running water after exposure to sodium sulfite solutions. See section 7.6.

## 8.0 Quality Control

Samples are done in duplicate. The Winkler method is used to calibrate the oxygen electrode.

## 9.0 Data Analysis

See sections 7.1.2 and 7.2.5.

## 10.0 Documentation

See sections 7.2.1, 7.2.3, 7.2.7, and 7.3.9.

## 11.0 Records

See sections 7.2.7 and 7.3.9.